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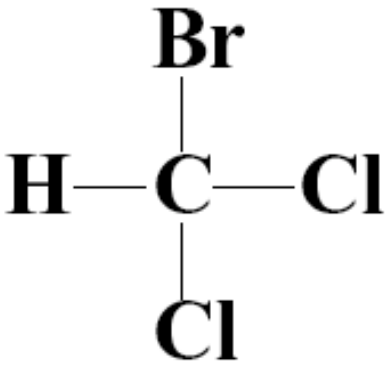
Toxicology & Carcinogenicity Studies of Bromodichloromethane



BROMODICHLOROMETHANE

CAS No. 75-27-4

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Nomination and Selection

Nomination

- BDCM was nominated by EPA and NIEHS

Rationale

- Trihalomethanes are some of the most common DBPs found in municipal drinking water. Colon cancer has been associated with BDCM gavage exposure in rats but not when BDCM is given in the drinking water
- Genetically modified mouse models may provide mechanistic insights into route differences in cancer response.

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DBP Selection

Dichloroacetic acid (DCA)

- DCA is well studied rodent liver carcinogen
- Representative haloacetic acid second most common DBP
- Weakly mutagenic

Bromodichloromethane (BDCM)

- BDCM is well studied rodent carcinogen (colon, kidney - rats, kidney - male mice, liver - female mice).
- Representative trihalomethane
- Mutagenic in bacterial systems, does not induce micronuclei

Bromate

- Common anion found after ozonation of water
- Well studied renal carcinogen in rodents, mutagenic

Study Summary Tg.AC Mouse Model

6- and 9-month[a] Dermal Studies

- Dermal administration 5 days/week
- Doses: 64, 128 and 256 mg/kg + vehicle control

6- and 9-month Drinking Water Studies

- Animals: male and female mice
- Concentration in water: 0, 175, 350, 700 mg/L

6- and 9-month Gavage Studies

- Animals: male and female mice
- 25, 50, 100 mg/kg + Corn Oil Vehicle

[a]Dermal exposure was for 26 & 39 weeks

Survival and Body Weights in the BDCM Tg. AC Dermal Studies

6-month Studies

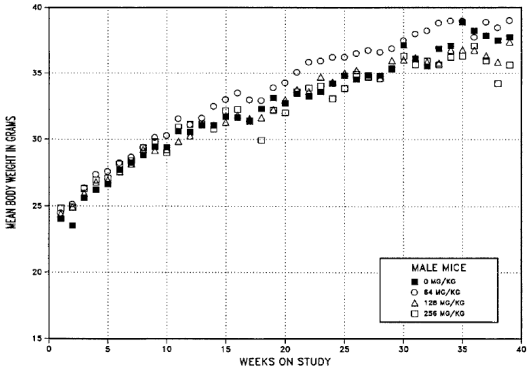
- Males — No effect on body weight or survival
- Females — No effect on body weight or survival

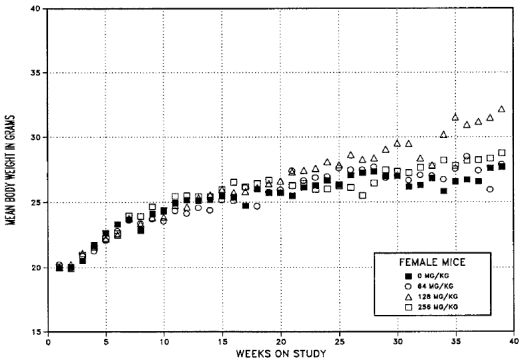
9-month Studies

- Males — No effect on body weight or survival
- Females — No effect on body weight or survival

No significant increase in neoplastic or nonneoplastic lesions at either 6 or 9 months in mice exposed to 64, 128 and 256 mg/kg

Males
9 mo.





Females
9 mo.

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Study Summary Tg.AC Mouse Model

6- and 9-month Dermal Studies

- Doses: 64, 128 and 256 mg/kg + vehicle control

6- and 9-month[a] Drinking Water Studies at 0, 175, 350, 700 mg/L

- Dose to males (approximately 18, 33 or 64 mg/kg)
- Dose to females (approximately 28, 49 or 111 mg/kg)

6- and 9-month Gavage Studies

- Animals: male and female mice
- 25, 50, 100 mg/kg + Corn Oil Vehicle

[a]Drinking water exposure was for 26 & 42 weeks

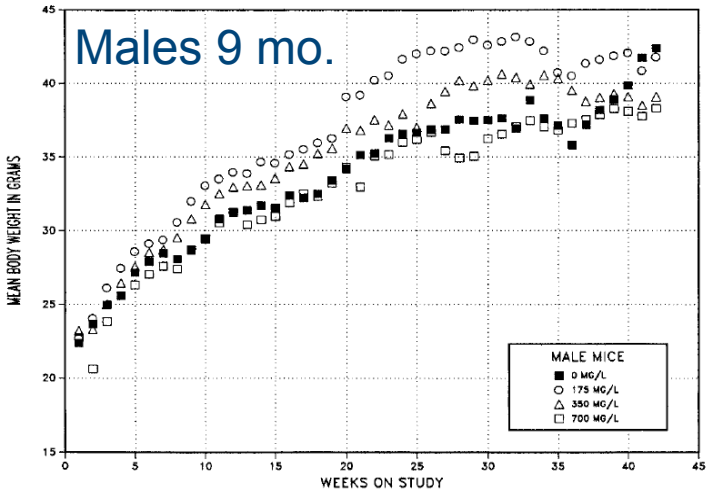
Survival and Body Weights in BDCM Tg. AC Drinking Water Studies

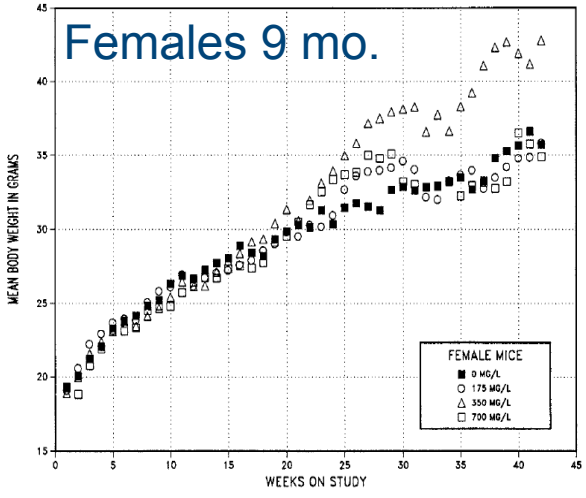
6-month Studies

- Males — No effect on survival; body weights decreased
- Females — No effect on survival; body weights increased

9-month Studies

- Males — Slight increase in body weight; no effect on survival
- Females — Slight increase in body weight; no effect on survival





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Renal (Male) & Hepatic (Female) Lesions in Tg.AC Drinking Water Study

Concentration (mg/L)	0	175	350	700
Male				
Nephropathy				
6-month[a]	27% (1.0)[c]	20% (1.3)	27% (1.3)	73%** (1.3)
9-month[b]	40% (1.0)	70% (1.0)	80% (1.3)	90%* (1.6)
Renal Tubule Degeneration				
6-month	0	27%* (1.0)	27%* (1.0)	60%**(1.3)
9-month	0	0	20% (2.0)	60%**(1.7)
Female				
Fatty Change				
6-month[a]	0	27%* (1.0)	53%** (1.1)	67%** (1.5)
9-month[b]	0	60%** (1.2)	60%* (2.0)	60%* (2.7)
Hypertrophy				
6-month	7% (2.0)	13% (2.5)	53%** (2.4)	80%** (2.8)
9-month	0	0	0	0

[a]N=15
[b]N=10
[c]Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked.
*P≤0.05, **P≤0.01

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Study Summary Tg.AC Mouse Model

6- and 9-month Dermal Studies

- Doses: 64, 128 and 256 mg/kg + vehicle control

6- and 9-month Drinking Water Studies at 0, 175, 350, 700 mg/L

- Dose to males (18, 33 or 64 mg/kg)
- Females (28, 49 or 111 mg/kg)

6- and 9-month[a] Gavage Studies

- Gavage 5 times/week to male and female mice
- 25, 50, 100 mg/kg + Corn Oil Vehicle

[a]Gavage exposure was for 26 & 41 weeks

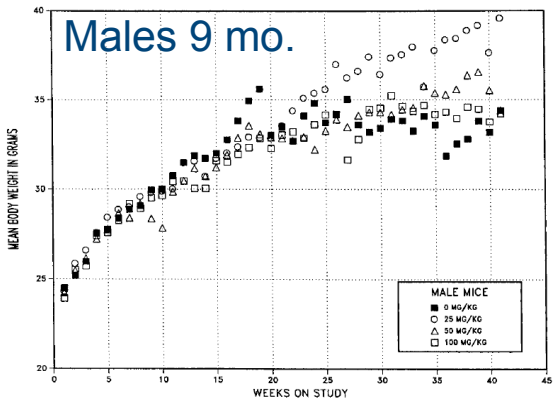
Survival and Body Weights in the BDCM Tg. AC Gavage Studies

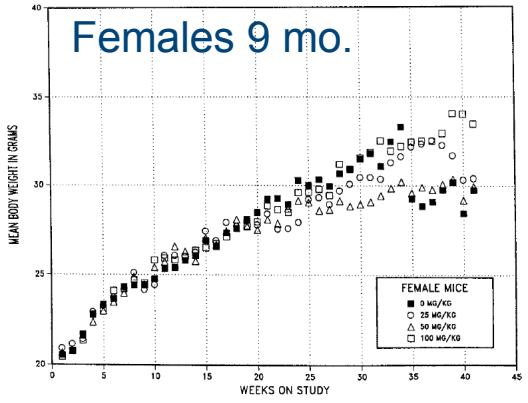
6-month Studies

- Males — No effect on survival; no decrease in body weight
- Females — No effect on survival; no decrease in body weight

9-month Studies

- Males — No effect on survival; no decrease in body weight
- Females — No effect on survival; no decrease in body weight





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Neoplastic/Nonneoplastic Lesions in Tg.AC Gavage Study

Dose (mg/kg)	0	25	50	100
Male				
Forestomach Pap.[a]				
6-month[b]	60%	33%	73%	80%
9-month[c]	80%	60%	90%	60%
Renal Tubule Degeneration				
6-month	0	0	0	27%(1.0)[d]
9-month	0	0	0	60%**(1.7)
Female				
Forestomach Pap.[a]				
6-month[b]	40%	53%	67%	73%
9-month[c]	40%	70%	80%	100%**
Fatty Change				
6-month	0	33%* (1.0)	53%** (1.0)	47%** (1.1)
9-month	0	20% (1.5)	80%** (1.4)	50%* (1.6)

[a] Includes all papillomas
[b] N=15
[c] N=10
[d] Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked.
*P≤0.05, **P≤0.01

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BDCM Conclusions

Male and Female Tg.AC mice

No increased incidence of neoplasms in male and female mice exposed to BDCM either dermally or in the drinking water or in males exposed by gavage.

Increased incidence of multiple forestomach papillomas in female mice exposed to BDCM by gavage for 6 or 9 months.

Increased incidences and/or severities of renal tubular degeneration in male mice and fatty change and/or hepatocyte cytoplasmic vacuolization in females exposed to BDCM in the drinking water or by gavage.

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Study Summary p53 Mouse Model

6- and 9-month Drinking Water Studies[a]

- Concentration in water: 0, 175, 350, 700 mg/L
  - Approximately 0, 14, 30 and 55 mg/kg males
  - Approximately 0, 22, 43 and 98 mg/kg females

6- and 9-month Gavage Studies[b]

- Animals: male and female mice
- 25, 50, 100 mg/kg + Corn Oil Vehicle

[a]Drinking water exposure was for 26 & 42 weeks

[b]Gavage exposure was 5 times/week for 26 & 41 weeks

Survival and Body Weights in the BDCM p53 Drinking Water Studies

6-month Studies

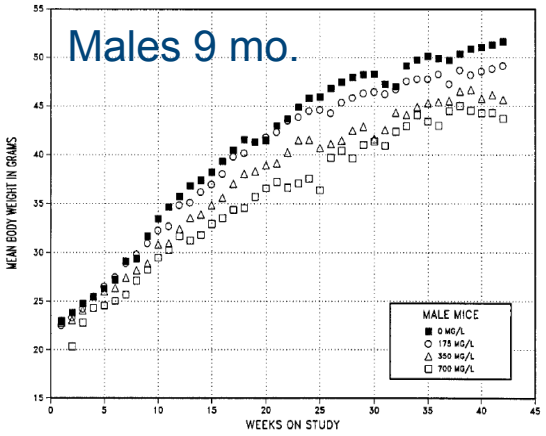
- Males — No effect on survival; decreased body weight
- Females — No effect on survival; decreased body weight

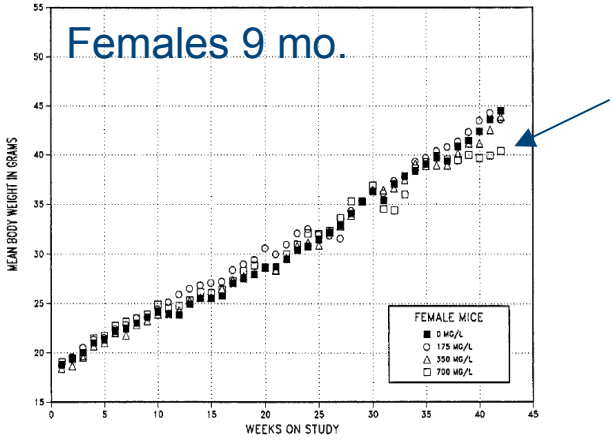
9-month Studies

- Males — No effect on survival; decreased body weight
- Females — No effect on survival; decreased body weight

Water consumption (1st 13 wks) : Males 50% of controls

Females approximately 70%





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Renal (Male) & Hepatic (Female) Lesions in BDCM p53 Drinking Water Study

Concentration (mg/L)	0	175	350	700
Male				
Nephropathy				
6-month[a]	47% (1.0)[c]	20% (1.0)	73% (1.5)	87%* (2.1)
9-month[b]	50% (1.0)	40% (1.0)	90%* (1.2)	80%* (1.4)
Renal Tubule Degeneration				
6-month	0	0	60%** (1.0)	80%* (1.0)
9-month	0	0	60%** (2.2)	100%**(2.4)
Female				
Fatty Change				
6-month[a]	0	7% (1.0)	7% (1.1)	67%** (1.0)
9-month[b]	20% (1.0)	20% (1.0)	30% (1.0)	60%* (1.7)

[a]N=15
[b]N=10
[c]Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked.
*P≤0.05, **P≤0.01

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Study Summary p53 Mouse Model

6- and 9-month Drinking Water Studies[a]

- Concentration in water: 0, 175, 350, 700 mg/L
  - Approximately 0, 14, 30 and 55 mg/kg males
  - Approximately 0, 22, 43 and 98 mg/kg males

6- and 9-month Gavage Studies[b]

- 25, 50, 100 mg/kg + Corn Oil Vehicle
- Gavage dose in males approximately 2-fold water study
- Gavage dose in females approximately equal water study

[a]**Drinking water exposure was for 26 & 42 weeks**

[b]**Gavage exposure was 5 times/week for 26 & 41 weeks**

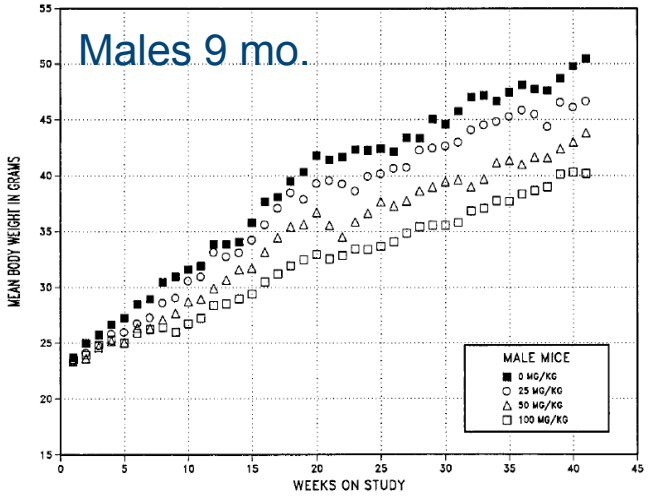
Survival and Body Weights in the BDCM p53 Gavage Studies

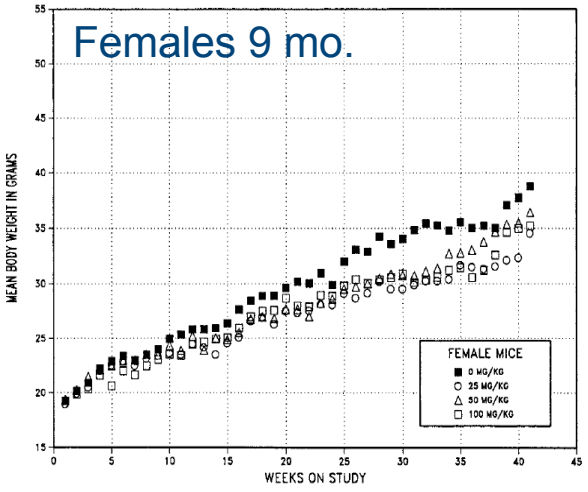
6-month Studies

- Males — No effect on survival; decreased body weight
- Females — No effect on survival; decreased body weight

9-month Studies

- Males — No effect on survival; decreased body weight
- Females — No effect on survival; decreased body weight





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Renal (Male) & Hepatic (Female) Lesions in BDCM p53 Gavage Study

Dose (mg/kg)	0	25	50	100
Male				
Nephropathy				
6-month[a]	53% (1.0)[c]	60% (1.0)	53% (1.0)	53% (1.1)
9-month[b]	40% (1.0)	30% (1.0)	40% (1.0)	90%* (1.0)
Renal Tubule Degeneration				
6-month	0	0	0	27%* (1.3)
9-month	0	10%(3.0)	0	100%**(2.5)
Female				
Fatty Change				
6-month[a]	13% (1.0)	13% (1.0)	20% (1.1)	73%** (1.3)
9-month[b]	30% (1.3)	30% (1.3)	60% (1.3)	90%** (2.3)

[a]N=15
[b]N=10
[c]Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked.
*P≤0.05, **P≤0.01

BDCM Conclusions

Male and Female p53 haploinsufficient mice:

No evidence of carcinogenic activity:

175, 350 and 700 mg/L in drinking water for 6 or 9 months

25, 50 and 100 mg/kg by gavage 5 x/week for 6 or 9 months

- Increased incidence and/or severities of renal tubule degeneration in males by both routes
- Increased incidence and/or severities of hepatocyte fatty change in females by both routes

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Conclusions on BDCM in GMM Models

1) BDCM caused forestomach papillomas in Tg.AC females

1) Forestomach response by gavage **only** is consistent with prior studies
2) Six month exposure gave suggestive forestomach effect

2) Rodent carcinogen failed to cause cancer in Tg.AC mouse by dermal and drinking water routes & in males by gavage

3) Rodent carcinogen failed to cause cancer in p53 model

Decreased body weight suggest higher dose not possible

1. No response by both drinking water and gavage routes
2. No insight on gavage carcinogenic response in rats/mice
3. BDCM is weakly mutagenic

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NTP Technical Reports Review Subcommittee Meeting

Bromodichloromethane

GMM 5